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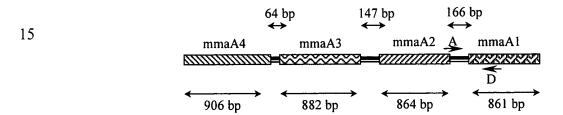


Figure 1: Schematic diagram of methoxy mycolic acid synthase mmaA 4-mmaA 1 gene cluster of mycobacteria and location of forward A, and reverse D primers.

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| ICAACCATTTCAATCGTCGGAGGTTGGCCCCCGGGAAGATTTCGGTCGCGATGAACTTGAGAAAGCGGGCCAGCC CCTAGCTTGCCCAGCGCCAGGTCGATCTTGGCGATCTGGGCCTCTTCCAGCGTCATGTCCTCGCGTTCGAAATGCGC ICTTCGGACTGTACGGCGATGGCGCGTTCGCGGGCAGCCTGTAGGTTGGCGGCCCATGCATC*GAGAGTCCGTGCGTA* ICTITIGCTGCGTTCATAGTGATTCCGGCTGAGCGTGAGGCCGATGACATTGACGTCGTACTTCTCCACGGCCCGAAC ATGTACCGCTCGTAGACCTCTTCGGACTGGATCGCGATGGCCTCGCTTTTGTGTTCCTGCAGCGCCTCGGCCCACAG GICGAGGGICCTGGCGTAATGCGGCTGCAGCGACTGGCGGCGAGTCAGCGTGAAACCCGTCTTCGCCGACTGTTCC GAGCGCCCCGCCCCACCCACCTCGAGTAGCGTCATCCCCGGTTCGAGGTTCAGCTTGTCCAACGCCAGA GATCGAGGAACAACGCGAAGAAGTCATCCGAAATGTCGTAAGCCGACTGTGACTCTTCGTAATATGGTCTCAGCTT 2GATCGACACGATGCGGTCGACGGGCTCGTTGAACTGCTCCCATCCCGCCAGCAACACTCGCCTGTCGCGGGGGT gcatgctcgaggcatgctatccgatacagggccgcactaaaccgcgatcgaatttgcccaggtcagggaacggatatgagcggacgacgagCTACTTGGTCATGGTGAA $g_{\overline{I}\overline{G}}$ CGTAGGAGCGTTCGAAGAACGTCAGATACCGCTCCTTTTTGAACGCCTCGAATGCCTCAAAGCTGACGATCCGGTC CTACTTCGCCAGCGTGAACTGGTTGACGTCGATGTAGCCGACCCGGAACAGCTTGGCGCAGCCGGTCAGGTATTTC CTGGGCGACGTTGATTAGGCCTCTGCGGAAGCGCTCCGCCATCCGGTCAGATAGTGCATGAAGTTGTTGTAGACC ACAACGTGAGCGGCAAGCCGTGGTCGACCATCTGCTGCTGGTCAGGCCGGTGATCGTGTGCAGCAGCACACGC CATCGGGCGGCAGGATTTTGTGGGCCCGGGCGAAGAAGTCGGCGTGACGATCGTGGCCCGAAGTGCTCGAACGCGC GCAGCTGTAGGTCTGGGTCCGGATCCAGGAACAGCCGGAAGAAGAGTCGTCGGACAGGTCGTAGTGTGCCTGCACGTC CTCGAAGTGCGGCGTTAGGTCGTTGACCATgaggtgtaatgcctttccggaccctaggtggcctttcggtgcttgcacggaacgcaccgatgcttcccctcccc GGCAGCTCGCCCGGGGAAGATCGACTCCCGCAGGAATTTGAGGAATCGAAGGTCGCTCATCGTCAGCGCAATG CCCTGTTCGTGCAGCCACCTGCGGTCGTAGGTGAACAGGCTGTGCAGTAGCATCCGCCCGTCATCGGGCAGGATGT GACGTTCTTCAAACTCTTCCCAGCCCTGCAGCCGGGCCTCGGCGCGCCGTTGCGTTCCGATTGCGGCCAGGCGG TCCACCTTGGCCAGTTGCGCCTCTTCCAGCGTCATATCGTCACGCTCGAAATAGGCGCAGGTGTAGACCCAGGTGG STCCATCTCGTCGAACGACTTCTGCACATGGGCGGCCTGGTTCTTCGACAATGTCAGGCCGACGACGTTGACGTCA
 FACTGCGCGCGCGCGCCGCATGGTGGCGCCCCAGCCGCAACCGATATCGAGCAGCGTCATGCCGGGCTGCAGA

Fig. 2: Sequence of mmaA2 and mmaA1 gene with an intergenic region of 166 base pair (shown in lower case. Location of forward A, sequence ID 1 and reverse primer D, sequence ID 2. Both primer sequence is underlined and italisized.

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1 2 3 4 5 6 7 8 9 10 11 12 13 14

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Fig. 3 PCR amplification of different mycobacterial genomic DNAs with primers A and D (lanes 1- 15): 1. M.avium 2. M.bovis 3. M.chelonae 4. M.fortuitum 5. M.intracellulare 6. M.kansassi 7. M.phlei 8. 100 bp DNA ladder 9. M.marinum 10. M.scrofulaceum 11.M.smegmatis 12. M.szulgai, 13. M.tuberculosis and 14. negative control. AD indicates 363 bp-amplified product.

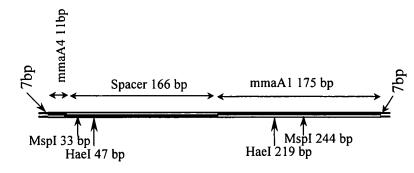


Fig. 4: line diagram showing restriction endonuclease map of HaeI and MspI within AD.

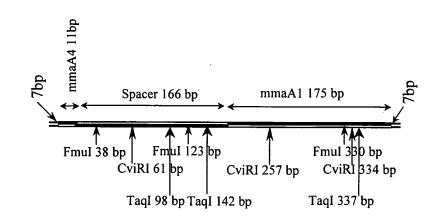


Fig. 5: line diagram showing restriction endonuclease map of Fmul, CviRI and TaqI within AD.



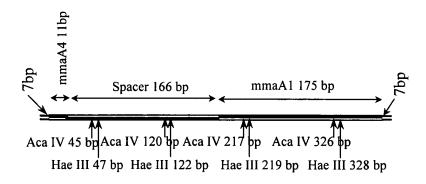


Fig. 6: Restriction map of AD showing distribution of the sites of restriction endonucleases AcaIV and HaeIII.

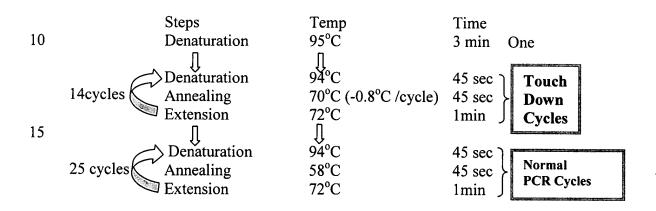


Fig. 7: Line diagram showing different steps of PCR reaction